09936869 . 032902

#### REMARKS

Obvious inadvertent clerical and typographical errors in the Specification are corrected in the present Amendment, and a paragraph providing cross reference to related applications has been inserted. Certain claims have been amended to eliminate multiple dependencies. None of the amendments made herein constitutes the addition of new matter.

It is believed that this Amendment does not necessitate the payment of any fees under 37 C.F.R. 1.16-1.17. If this is incorrect, please deduct any fee due under the foregoing Rules from Deposit Account No. 07-1969.

Respectfully submitted,

Donna M. Ferber Reg.No. 33,878

GREENLEE, WINNER AND SULLIVAN, P.C. 5370 Manhattan Circle, Suite 201 Boulder, CO 80303 Telephone (303) 499-8080

Facsimile: (303) 499-8089 Email: winner@greenwin.com

Attorney docket No.103-01 bmk:March 29, 2002



# Marked up version of amended claims and paragraphs in attached Amendment.

Docket No.: 103-01 USSN 09/936,869

Filed: September 17, 2001

## At page 4, the paragraphs describing Figures 1 and 2:

Figure 1 shows the amino acid sequence of the reproductive peptide PrAG1, together with the nucleotide sequence coding therefor (SEQ ID NO:3);

Figure 2 shows the sequence of the PrAG1 promoter, which is the focus of the present invention, isolated from *Pinus radiata* (SEQ ID NO:1);

## At page 14, the sequences beginning at line 28:

3' PCR primer: 5' GCIGTIAGIYCITCICCCAT 3';

(SEQ ID NO:[6]<u>7</u>)

5' PCR primer: 5' AAYCGICARGTIACITT 3'

(SEQ ID NO:[7]8)

## At page 16, the sequences beginning at line 31:

Primer GSP1: 5' CGC CTT CTT CAA TAA ACC ATT TCG GCG CTT 3' (SEQ ID NO:[8]9)

Primer GSP2: 5' GAC CTG TCG GTT CGT AGT ATT TTC AAT CCT 3' (SEQ ID NO:[9]10)

### At page 17, lines 4 and 5:

Primer GSP3: 5' TTC GTC CTC CAT TTT GTG CGC TCT CCA TTC 3' (SEQ ID NO:[10]11)

Primer GSP4: 5' GCA CTC CAC TCT TCC TTT ATT TCT TAC CAC 3' (SEQ ID NO:[11]12)

#### At page 19, the paragraph beginning at line 18:

Analysis was performed on total RNA isolated from needle, stem, vegetative shoot, immature female cone and immature male cone samples as described above. RNA was reverse-transcribed with MMLV reverse-transcriptase (Gibco BRL) according to the manufacturer's instructions. PCR was performed with two primers: 5'PCR primer (5' TTGTGTACAAATCATGGG 3') (SEQ ID NO:[13]14) and 3'PCR primer (5'GTAAGCCCGTCACCCATC 3') SEQ ID NO:[14]15). Verification of the specificity of the PCR reactions was achieved through the use of controls that

included amplification reaction with single primers, RNAse treatment of template, and no template. In those reactions in which no PCR product was detected, the quality of the RNA was tested by UV scanning and agarose gel electrophoresis. ss-cDNA from the RTR reaction was used as a template. The 50-ul reaction mixture contained 2.5 U Taq DNA [polymease] polymerase, 1X Polymerization Buffer (both from ClonTech Co.), 1mM MgCl<sub>2</sub>, 0.2mM dNTP and 0.25  $\mu$ M primers. The PCR was performed under following conditions: denaturation at 94°C for 30 s, annealing at 55°C for 1 min and extension at 72°C for 1 min for 30 cycles on Thermal Cycler 480 (Perkin-Elmer, Norwalk, CT, USA). The PCR products were subjected to electrophoresis in agarose gel, and hybridization as described above.

#### At page 21, the paragraph beginning at line 7:

To check the genomic DNA integration of pRAGPR in the transgenic tobacco plants, gene-specific primers for the NPTII gene were employed. The primers used were NPTII-5' primer 5-GAA CAA GAT GCA TTG CAC GC-3' (SEQ ID NO:[15]16) and NPTII-3' primer 5'-GAA GAA CTC GTC AAG AAG GC-3' (SEQ ID NO:[16]17). Genomic DNA from each of the control lines and transgenic tobacco lines were isolated from the leaf tissue using the Qiagen DNAeasy kit as per manufacturer's instructions. PCR reactions (50-μl final volume) were performed using 5 μl of template DNA. Samples were heated to 95°C for 4 minutes, followed by 35 cycles of 95°C for 45 seconds, 55°C for 30 seconds, and 73°C for 2 minutes, with a final extension step of 73°C for 5 minutes in PTC100 thermal cycler (MJ Research). Amplified DNA fragments were analyzed on a 0.8% agarose gel and visualized by staining with ethidium bromide.

#### In the Claims:

- 5. (Twice amended) A DNA construct which comprises:
  - (a) a promoter sequence [according to claim 2 or claim 3] as given in SEQ ID NO:1 or a functionally equivalent variant thereof which has at least 90% homology to SEQ ID NO:1 or as given in SEQ ID NO:2;
  - (b) an open reading frame polynucleotide coding for a peptide; and
  - (c) a termination sequence.

- 9. (Once amended) A construct according to [any one of] claim[s 4-7] 6 wherein said open reading frame polynucleotide encodes a peptide which, when expressed in reproductive tissue of a plant, causes said plant's reproductive organs to abort.
- 10. (Once amended) A construct according to [any one of] claim[s 4-7] 6 wherein said open reading frame polynucleotide encodes a peptide which, when expressed in reproductive tissue of a plant, causes said plant's reproductive organs to redefine themselves as vegetative.
- 11. (Once amended) A construct according to [any one of] claim[s 4-7] 6 wherein said open reading frame polynucleotide encodes a peptide which, when expressed in reproductive tissue of a plant, causes said plant's reproductive organs to stop development.
- 12. (Once amended) A construct according to [any one of] claim[s 4-7] 6 wherein said open reading frame polynucleotide encodes a peptide which, when expressed in reproductive tissue of a plant, causes cell death.
- 16. (Once amended) A construct according to [any one of] claim[s 4-7] 6 wherein said open reading frame polynucleotide encodes a peptide which, when expressed in reproductive tissue of a plant, causes an alteration in the timing of flowering of said plant.
- 17. (Once amended) A construct according to [any one of] claim[s 4-16] 5 which further includes:
  - (d) a selection marker sequence.
- 19. (Once amended) A transgenic plant cell which includes a construct according to [any one of] claim[s 4-18] 5.
- 20. (Once amended) A transgenic plant which includes a construct according to [any one of] claim[s 4-18] 5.

- 21. (Once amended) A transgenic plant which contains a polynucleotide according to claim 1 or a promoter according to claim [2 or 3] 5, which plant has a reduced reproductive capacity.
- 25. (Once amended) A transgenic plant according to [any one of] claim[s 20-24] 20 wherein said plant is a coniferous plant.
- 28. (Once amended) A transgenic plant according to [any one of] claim[s 20-24] <u>20</u> wherein said plant is a tree.